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A CHEMICAL LURE FOR STINK BUGS (HEMIPTERA: PENTATOMIDAE) IS USED AS A KAIROMONE BY *ASTATA OCCIDENTALIS* (HYMENOPTERA: SPHECIDAE)

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ABSTRACT

We tested lures in Washington and Georgia containing methyl (*E,E,Z*)-2,4,6-decatrienoate and/or methyl (*E,Z*)-2,4-decadienoate for capturing stink bug species (Hemiptera: Pentatomidae). In both states, we consistently captured adult females of the digger wasp *Astata occidentalis* Cresson (Hymenoptera: Sphecidae) in traps with lures that contained methyl (*E,E,Z*)-2,4,6-decatrienoate but not in traps with lures that contained only methyl (*E,Z*)-2,4-decadienoate. *Astata occidentalis* is a predator of pentatomid stink bugs (Hemiptera) and apparently uses methyl (*E,E,Z*)-2,4,6-decatrienoate as a host-finding kairomone. Methyl (*E,E,Z*)-2,4,6-decatrienoate is an aggregation pheromone of *Plautia stali* Scott (Hemiptera: Pentatomidae) that is also attractive to, and used for monitoring, the exotic brown marmorated stink bug, *Halyomorpha halys* Stål (Hemiptera: Pentatomidae). Wasps were captured from Jul through Sep with peak response in Aug in Washington. In Georgia, wasp capture was highest as soon as traps were deployed in late May/early Jun, and then capture peaked again during Jul.

Key Words: *Halyomorpha*, digger wasp, trap, lure, pheromone, kairomone

RESUMEN

En los estados de Washington y Georgia, probamos señuelos que contienen (*E,E,Z*)-2-4-6-decatrienoato de metilo y/o (*E,Z*)-2-4-6-decatrienoato de metilo para la captura de especies de chinches hediondas (Hemiptera: Pentatomidae). En ambos estados, capturamos consistentemente hembras adultas de la avispa excavadora *Astata occidentalis* Cresson (Hymenoptera: Sphecidae) en trampas con señuelos de (*E,E,Z*)-2-4-6-decatrienoato de metilo pero no en las trampas con señuelos de sólo (*E,Z*)-2-4-6-decatrienoato de metilo. *Astata occidentalis* es un depredador de chinches pentatomidas (Hemiptera) que aparentemente utiliza el (*E,E,Z*)-2-4-6-decatrienoato de metilo como una kairomona para encontrar su hospedero. El (*E,E,Z*)-2-4-6-decatrienoato de metilo es una feromonía de agregación de *Plautia stali* Scott (Hemiptera: Pentatomidae) que también es atractivos y utilizado para el monitoreo del chinche exótico marmorado, *Halyomorpha halys* Stål (Hemiptera: Pentatomidae). Las avispas fueron capturadas desde julio hasta septiembre, con la respuesta más alta en agosto en el estado de Washington. En Georgia, la captura de avispas obtuvo un punto alto tan pronto como las trampas fueron puestas en de final del mes de mayo/principio de junio, y luego la captura alcanzó otro punto máximo de nuevo durante julio.

Palabras Clave: *Halyomorpha*, avispa excavadora, trampa, atractivo, feromonas, kairomona

Astata occidentalis Cresson (Hymenoptera: Sphecidae) has a broad distribution in the U.S. (Evans 1957). It is a predatory solitary wasp that provisions a subterranean nest with true bugs (i.e., Heteroptera) including the Pentatomidae (i.e., stink bugs) (Bohart & Menke 1976). It is

known to utilize stink bug species in several genera, including *Thyanta*, *Euschistus*, *Banasa*, and *Hymenarcys* sp. (Evans 1957; O'Neill 2001).

This wasp can apparently use the stink bug pheromone produced by *Thyanta pallidovirens* (Stål), i.e., methyl (*E,Z,Z*)-2,4,6-decatrienoate, as

a host-finding kairomone (Millar et al. 2001). Additionally, *A. occidentalis* broadly overlaps with the present distribution of the invasive brown marmorated stink bug, *Halyomorpha halys* (Stål) (Hemiptera: Pentatomidae) in the U.S. (Leskey et al. 2012a) and, interestingly, the *EEZ* isomer of methyl (*E,Z,Z*)-2,4,6-decatrienoate is an attractant for *H. halys* (Leskey et al 2012b).

Attractant traps are available for detection of the presence and relative abundance of some stink bug species including *Euschistus* spp. (Aldrich et al. 1991) and *H. halys* (Leskey et al. 2012b). The male-specific pheromone of *Euschistus* spp., i.e., methyl (*E,Z*)-2,4-decadienoate, attracts males, females, and nymphs of *E. servus* (Say) and other *Euschistus* spp. in the field. It also serves as a host-finding kairomone for parasitoids of *E. servus*, including *Gymnosoma* and *Euthera* spp. (Diptera: Tachinidae) (Aldrich et al. 1991, 2007). *Halyomorpha halys* were first trapped with methyl-(*E,E,Z*)-2,4,6-decatrienoate (Lee et al. 2002), which is an aggregation pheromone of the Oriental stink bug *Plautia stali* Scott (Sugie et al. 1996).

Our objective was to use field studies to evaluate *H. halys* responses to aggregation pheromones of *Euschistus* spp. stink bugs, i.e., methyl (*E,Z*)-2,4-decadienoate, and *P. stali*, i.e., methyl (*E,E,Z*)-2,4,6-decatrienoate, when used alone or when the two were combined. Only two *H. halys* males were captured, both in methyl (*E,E,Z*)-2,4,6-decatrienoate-baited traps in Washington, during this experiment. We did, however, consistently capture numbers of *A. occidentalis* in traps baited with methyl (*E,E,Z*)-2,4,6-decatrienoate at both locations. Thus, we report here the responses of *A. occidentalis* wasps to traps with lures containing methyl (*E,Z*)-2,4-decadienoate and/or methyl (*E,E,Z*)-2,4,6-decatrienoate.

MATERIALS AND METHODS

Trapping studies in Washington and Georgia, unless otherwise stated, used the RESCUE!® Stink Bug Trap baited with Rescue® lures (Sterling International, Spokane, Washington, USA) or polyethylene pouch dispensers (4 mil polyethylene, 2.1 × 5.5 cm), respectively, and each type of lure contained methyl (*E,E,Z*)-2,4,6-decatrienoate (Bedoukian Research Inc., Danbury, Connecticut, USA). Additional studies evaluated methyl (*E,Z*)-2,4-decadienoate (Bedoukian Research Inc., Danbury, Connecticut, USA).

The design of the RESCUE® Stink Bug Trap (<http://www.rescue.com/product/reusable-outdoor-stink-bug-trap>) consisted of a clear plastic 8-cm-diam cylinder (0.9 L) set over the top of 3 green vanes that narrow from bottom to top (i.e., a pyramidal shape) and led upward into a perforated, translucent plastic funnel set inside the cylinder (Schneidmiller et al. 2011). The diam-

eter of the large funnel opening, connecting to the vanes, was 7.5 cm, while the diam of the top, i.e., smaller funnel opening, was 1.0 cm. The length of the trap was 38 cm.

Trapping Tests in Washington

Nine RESCUE® Stink Bug Traps baited with RESCUE!® stink bug lures were placed in a variety of habitats in early Jul 2012, in the vicinity of Yakima, Washington. Each trap was at a different site, with sites from 100 m to several km apart. Trap site habitats included urban, suburban yards, rural agriculture, and riparian. Traps were placed at a height of 1.5 to 2.0 m in trees and shrubs. Traps were checked twice per wk, and pheromone lures were replaced on 11 Aug. Traps were maintained through Oct.

Trapping Tests in Georgia

Attractant Comparison Study. Sets of RESCUE® Stink Bug Traps were deployed at 2 locations during 2011 in Elbert County (near Elberton), McDuffie County (near Dearing), and Union County (near Blairsville), and at 4 locations in Peach County (2 sites at the USDA, ARS Laboratory near Byron, 1 site near Ft. Valley, and 1 site near Marshallville), all in Georgia. In addition to using methyl (*E,E,Z*)-2,4,6-decatrienoate, we also used the aggregation pheromone of *Euschistus* spp. stink bugs, i.e., methyl (*E,Z*)-2,4-decadienoate. Each location had 4 treatments with each set of traps: 1) 50 mg methyl (*E,E,Z*)-2,4,6-decatrienoate in a polyethylene pouch, 2) 100 mg methyl (*E,Z*)-2,4-decadienoate in a polyethylene pouch, 3) 50 mg of methyl (*E,E,Z*)-2,4,6-decatrienoate plus 100 mg methyl (*E,Z*)-2,4-decadienoate in polyethylene pouches and 4) control with no lure. Traps were hung ca 1.5 m above ground from tree branches, trellises, or fencing. Within the 4 treatments, traps were spaced 10 m apart. Where more than 1 set of traps was placed at a location, sets of traps were > 100 m apart. At all locations, traps were placed in the field on May 24, 2011 and monitored once per week until Aug 2, Jul 6, Jul 26, or Aug 16 for Elbert, McDuffie, Peach and Union County sites, respectively. Lures at all locations were replaced once, on Jun 28, 2011. At the 2 Union County locations, data on *A. occidentalis* were not collected during the first wk.

Attractants, Doses and Trap Type Study. This study was done at the USDA, ARS facility at Byron, Georgia using two separate experiments (one using the RESCUE!® Stink Bug Trap and the other using yellow pyramidal stink bug traps) evaluating the same attractant treatments. For these experiments, polyethylene pouch dispensers were loaded with different doses of methyl (*E,Z*)-2,4-decadienoate or methyl (*E,E,Z*)-2,4,6-decatrienoate.

The yellow pyramidal stink bug traps used were as described by Cottrell and Horton (2011). This trap consisted of a 2.84-L clear polyethylene terephthalate jar (United States Plastic Corp, Lima, Ohio, USA) on top of a 1.22-m-tall yellow pyramidal base formed by 4 vanes made of Masonite fiberboard (Mizell & Tedders 1995; Cottrell et al. 2000). An insecticidal ear tag (Saber™ Extra, Coppers Animal Health Inc., Kansas City, Kansas, USA) was placed in the plastic jar to kill captured insects (Cottrell 2001). The active ingredients in the ear tag were lambda-cyhalothrin (10%) and piperonyl butoxide (13%). These traps were secured to the ground within an area of closely mowed vegetation.

Eight treatments were used for each experiment: 30, 50, 100, 150, and 200 mg methyl (*E,Z*)-2,4-decadienoate, or 100 mg methyl (*E,Z*)-2,4-decadienoate + 50 mg of methyl (*E,E,Z*)-2,4,6-decatrienoate per polyethylene pouch (formulated by Sterling International Inc., Spokane, Washington). These attractants and doses were chosen for the purpose of testing an attractant lure system for capturing multiple stink bug species (native and invasive) in the same trap. As such, it was necessary to determine if positive (i.e., additive or synergistic) or negative (i.e., antagonistic) interactions existed between the two attractants. The two other trap treatments were 40 μ L of methyl (*E,Z*)-2,4-decadienoate loaded onto a rubber septum (11 mm sleeve stopper, natural red rubber, Wheaton Inc., Millville, New Jersey, USA) and a blank control. A randomized complete block design with six replicates was used. RESCUE® Stink Bug Traps were hung 1.5 m above ground from branches, traps within replicates were spaced 10 m apart, and experimental blocks (replicates) of traps were spaced $>$ 100 m apart. Habitats for these traps included a weedy field near a wood lot, woods edges along peach orchards, pecan orchard edge row alongside a peach orchard, pecan orchard interior and a pecan orchard edge row near a pine woods. Traps were checked once per week from 12 Jun to 26 Jul 2011. When yellow pyramidal traps were used in the study, the traps were positioned on the ground and all traps within a replicate block were spaced 30 m apart and replicates were $>$ 100 m apart. Habitats for pyramidal traps included a woods edge along a peach orchard and woods edges along pecan orchards. Traps were checked once per week from 17 Jun to 29 Jul 2011. Lures were not changed during this study.

Statistical Analysis

Due to differences in lengths of collection periods, the mean numbers of *A. occidentalis* per treatment for the duration of the test were determined at the different trapping sites in the attractant comparison study and submitted to analysis

of variance (ANOVA). When a significant treatment effect was detected ($P < 0.05$), mean separation was done using Tukey's Honestly Significant Difference (HSD) Test (JMP 2012, SAS Institute, Cary, NC). The cumulative numbers of *A. occidentalis* collected from dose studies using RESCUE!® traps or yellow pyramidal traps were submitted separately to ANOVA. Again, when the treatment effect was significant ($P < 0.05$), mean separation was done using Tukey's HSD Test.

RESULTS

Washington Trapping Test

A total of forty nine *A. occidentalis* (mean \pm SE = 5.4 ± 1.6 , range = 1 to 13) were captured in these traps. The first wasp trapped was recorded 11 Jul and the last one trapped was recorded 20 Sep. All traps captured *A. occidentalis* and all specimens were female. Peak capture in Yakima County, Washington was in mid-Aug (Fig. 1).

Georgia Trapping Test

Attractant Comparison Study. *Astata occidentalis* were captured at all sites in each county when lures contained methyl (*E,E,Z*)-2,4,6-decatrienoate. Patterns of trap capture in each county were relatively similar for traps with lures containing only methyl (*E,E,Z*)-2,4,6-decatrienoate, except that peak capture in Union County occurred later than in Elbert and Peach counties; no data were collected in McDuffie County after early Jul (Fig. 2). Mean capture (\pm SE) of *A. occidentalis* in traps with lures containing methyl (*E,E,Z*)-2,4,6-decatrienoate, whether alone or in combination with methyl (*E,Z*)-2,4-decadienoate, was significantly higher (2.7 ± 0.7 and 2.1 ± 0.5

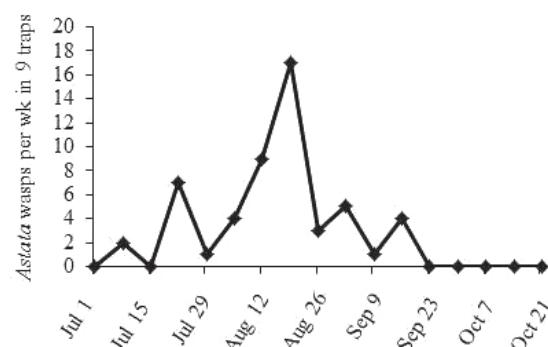


Fig. 1. Total numbers of *Astata occidentalis* wasps captured in RESCUE® Stink Bug Traps baited with RESCUE!® stink bug lures (Sterling International, Spokane, Washington, USA) for each week, from early Jul through Oct, 2012, in Yakima County, Washington.

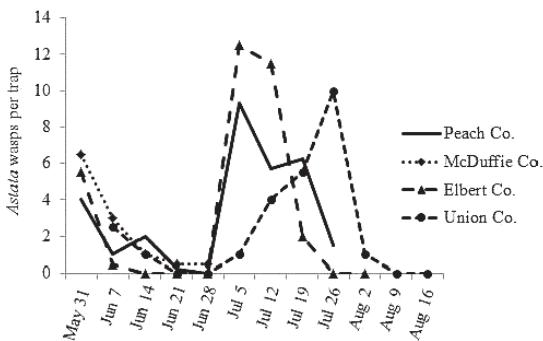


Fig. 2. Mean numbers of *Astata occidentalis* captured in 4 Georgia counties from late May through mid-Aug, 2011. Each Rescue® Stink Bug Traps was baited with a polyethylene pouch containing 50 mg methyl (*E,E,Z*)-2,4,6-decatrienoate.

per trap, respectively) than in traps with lures containing only methyl (*E,Z*)-2,4-decadienoate (0.36 ± 0.2 per trap) or in the control traps (0.0 ± 0.0) ($F = 8.79$; $df = 3, 27$; $P = 0.0003$).

Attractants, Doses and Trap Type Study. When methyl (*E,E,Z*)-2,4,6-decatrienoate was included in the lure, capture increased significantly over lures with any dose of only methyl (*E,Z*)-2,4-decadienoate in either RESCUE® Stink Bug Traps ($F = 12.48$; $df = 7,47$; $P < 0.0001$) or yellow pyramidal traps ($F = 6.11$; $df = 7,47$; $P = 0.0001$). RESCUE® Stink Bug Traps with pouch lures containing 0, 30, 50, 100, 150 or 200 mg of methyl (*E,Z*)-2,4-decadienoate captured an average (\pm SE) of 0, 0.2 ± 0.2 , 0.5 ± 0.2 , 1.3 ± 0.6 , 0.8 ± 0.7 and 4.0 ± 2.4 wasps per trap, respectively. The trap with the rubber septa lure, 40 μ l methyl (*E,Z*)-2,4-decadienoate, captured 1.2 ± 0.5 wasps per trap. In contrast, the trap that contained 100 mg of methyl (*E,Z*)-2,4-decadienoate + 50 mg of methyl (*E,E,Z*)-2,4,6-decatrienoate captured an average of 18.7 ± 4.8 wasps per trap. When the study was done using yellow pyramidal traps, the traps with pouch lures containing 0, 30, 50, 100, 150 or 200 mg of methyl (*E,Z*)-2,4-decadienoate captured an average of 0 , 3.3 ± 2.5 , 2.2 ± 1.4 , 3.7 ± 2.5 , 8.8 ± 4.0 and 3.3 ± 1.4 wasps per trap, respectively. The trap with the rubber septa lure, 40 μ l methyl (*E,Z*)-2,4-decadienoate, captured 2.3 ± 1.4 wasps per trap. For the yellow pyramidal trap with the lure containing 100 mg of methyl (*E,Z*)-2,4-decadienoate + 50 mg of methyl (*E,E,Z*)-2,4,6-decatrienoate an average of 103.8 ± 41.5 wasps were captured per trap.

DISCUSSION

This is the second record of *A. occidentalis* responding to a stink bug pheromone lure. Millar et al. (2001) captured the same predatory wasp,

A. occidentalis, in traps baited with the pheromone of the stink bug *Thyanta pallidovirens* (Stål). The pheromone of *T. pallidovirens* is methyl (*E,Z,Z*)-2,4,6-decatrienoate. Khrimian et al. (2008) report that methyl (*E,Z,Z*)-2,4,6-decatrienoate can isomerize, potentially producing other isomers, under UV exposure. So it is unclear at this time if *A. occidentalis* responded to the *EEZ* isomer in the lure or to some small amount of the *EZZ* isomer that may have been produced as the result of isomerization. However, far fewer *A. occidentalis* were trapped with methyl (*E,Z*)-2,4-decadienoate compared with methyl (*E,E,Z*)-2,4,6-decatrienoate.

Astata occidentalis were captured in each type of habitat sampled, and were captured throughout the sampling period. In Washington, this was from early Jul until mid-Sep. Rust et al. (1983) collected *A. occidentalis* during Jun, Jul and Aug in Nevada. In the current study, we did not have traps out before Jul and thus do not know when wasp activity begins, but no wasps were caught in traps in the field from mid-Sep until the traps were removed in December. In Georgia, wasps were captured throughout the entire lure testing period of late May to mid-Aug. Therefore we are unable at this time to make conclusions regarding the full seasonal activity period of *A. occidentalis* in either geographic area. Also, because pheromone lures were only changed once it is not possible for us to determine if changes in numbers of wasps captured were a direct result of changes in numbers of wasps present or were a result, at least in part, of pheromone depletion in lures. Although not compared directly, higher numbers of *A. occidentalis* were captured in yellow pyramidal traps than in RESCUE® Stink Bug Traps in Georgia. Overall, similar habitats were sampled by both trap types at the 465 ha research laboratory (USDA, ARS, SEFTNRL, Byron, Georgia) and any apparent differences would likely have been overcome by the mobility of the wasp. However, differences in trap design and color may have played a role. We do not present data regarding other natural enemies because none were captured consistently enough to provide a meaningful interpretation of the data.

There are other examples of predators and parasites responding to the pheromones of their prey. For example, the social wasp *Vespula germanica* (Fab.) locates male Mediterranean fruit flies, *Ceratitis capitata* (Wiedemann), by orienting to their pheromone (Hendrichs et al. 1994). Tachinid flies that are parasitoids of *Euschistus* stink bugs are attracted to the pheromone produced by male stink bugs (Aldrich et al. 1991, 2007).

Any role of *A. occidentalis* in the biological control of pest species of stink bugs is not known. Using kairomone-baited traps may be useful toward developing an understanding of the biology of this little-known predator.

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